

CLAIMS

1. A method for producing a nucleic acid product, comprising that a polymerase protein is contacted with a nucleic acid template under conditions sufficient for the function of the enzyme, and wherein said polymerase protein is an RNA polymerase capable of producing
 - (a) short complementary RNA copies of said template, which are scattered throughout the entire template length and, optionally,
 - (b) template-length complementary RNA copies.
2. The method according to claim 1, wherein said nucleic acid template is DNA or RNA.
3. The method according to claim 1, wherein the ratio of said short and template-length RNA copies can be adjusted by the reaction conditions.
4. The method according to claim 1, wherein the length of said short RNA copies can be adjusted by the reaction conditions.
5. The method according to claim 1, wherein the said short or template-length RNA copies are annealed to the template or denatured from the template.
6. The method according to claim 1, wherein said nucleic acid template is linear or circular.
7. The method according to claim 1, wherein said polymerase originates from a eukaryotic cell.
8. The method according to claim 7, wherein said polymerase originates from an organism selected from the kingdoms of Fungi, Viridiplantae, Metazoa, or the group of Mycetozoa.

9. The method according to claims 7 or 8, wherein said polymerase originates from an organism selected from the subset of genera *Neurospora*, *Arabidopsis*, *Caenorhabditis*, and *Dictyostelium*, preferably organisms *Neurospora crassa*, *Arabidopsis thaliana*, *Caenorhabditis elegans*, and *Dictyostelium discoideum*.

10. The method according to claim 9, wherein said polymerase is QDE-1 protein of *Neurospora crassa* or an altered or a genetically modified derivative of QDE-1.

11. The method according to any one of the preceding claims, wherein said RNA polymerase is encoded by a nucleic acid sequence selected from the group of:

(a) a nucleic acid sequence comprising the nucleic acid sequence of SEQ ID NO: 1 or SEQ ID NO: 3;

(b) a nucleic acid sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO 2 or SEQ ID NO: 4;

(c) a nucleic acid sequence, which differs from the nucleic acid sequence of (a) or (b) due to degeneracy of the genetic code;

(d) a nucleic acid sequence hybridizing to the nucleic acid sequence of (a), (b) and/or (c) under stringent conditions; and

(e) a nucleic acid sequence encoding a polypeptide comprising the amino acids 709 to 1402 of SEQ ID NO:4 or any sequence longer than that up to the sequence comprising the amino acids 2 to 1402 of SEQ ID NO: 2; and

(f) a nucleic acid sequence encoding an amino acid sequence, which shows at least 50% identity to the amino acid sequence of SEQ ID NO: 2.

12. An isolated polypeptide, characterized in that:

(i) said polypeptide has sufficient RNA polymerase activity;

(ii) said polypeptide has enhanced solubility resulting in at least 3 times higher yield of the active polymerase, than in the case of polypeptide comprising the amino acid sequence of SEQ ID NO: 2 or encoded by the nucleic acid sequence comprising SEQ ID NO: 1; and

(iii) said polypeptide is encoded by a nucleic acid sequence selected from the group of:

(a) a nucleic acid sequence comprising the nucleic acid sequence of SEQ ID NO: 3;

(b) a nucleic acid sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO: 4;

(c) a nucleic acid sequence, which differs from the nucleic acid sequence of (a) or (b) due to degeneracy of the genetic code;

(d) a nucleic acid sequence hybridizing to the nucleic acid sequence of (a), (b) and/or (c) under stringent conditions;

(e) a nucleic acid sequence encoding a polypeptide comprising the amino acids 709 to 1402 of SEQ ID NO:4 or any sequence longer than that up to the sequence comprising the amino acids 2 to 1402 of SEQ ID NO: 2; and

(f) a nucleic acid sequence encoding an amino acid sequence, which shows at least 50% identity to the amino acid sequence SEQ ID NO: 2.

13. The polypeptide according to claim 12, wherein said polypeptide has aspartate at the position 1011 of the amino acid sequence SEQ ID NO: 2.

14. The polypeptide according to claim 12, wherein said polypeptide is not encoded by a nucleic acid sequence encoding the amino acids 1 to 1402 or 710 to 1402 of the amino acid sequence of SEQ ID NO:2.

15. An isolated nucleic acid sequence encoding the polypeptide according to claim 12.

16. A vector, which comprises the nucleic acid sequence of claim 15 operationally linked with regulatory sequences required for gene expression.

17. A host cell comprising the vector of claim 15.

18. A method for producing a polymerase protein, which comprises culturing the host cell of claim 17 under conditions suitable for the expression of the protein.

19. The method according to claim 18, comprising the step that the protein is recovered from the cell or culture medium and optionally purified.

20. A method for producing RNA *in vitro*, comprising the steps of:

- (a) providing ssRNA or ssDNA template;
- (b) contacting said ssRNA or ssDNA template with the protein or polypeptide defined in claim 1 or 12 under conditions sufficient for RNA synthesis.

21. The method according to claim 20, wherein said ssRNA template is provided by transcribing a DNA template with a DNA-dependent RNA polymerase, preferably polymerase selected from the group of DNA bacteriophage-encoded DNA-dependent RNA polymerases, most preferably DNA-dependent RNA polymerase of bacteriophage T7, T3 or SP6.

22. The method according to claims 20 or 21, wherein steps (a) and (b) are carried out at the same time or sequentially in the same reaction vessel.

23. The method according to any one of claims 20 to 22, wherein the newly produced RNA species are recovered from the reaction mixture.

24. The method according to any one of claims 20 to 23, wherein said newly produced RNA strands are annealed with the template to form dsRNA elements or, alternatively, are denatured from the template.

25. The method according to any one of claims 20 to 24, wherein RNA synthesis is initiated from the 3' end of a nucleic acid primer complementary to the RNA or DNA template or RNA synthesis is initiated without a primer.

26. The method according to any one of claims 20 to 25, wherein the reaction mixture for RNA synthesis comprises at least one nucleoside triphosphate optionally labeled with a radioactive isotope or is chemically modified, pH buffer, ammonium acetate, PEG, Mg^{2+} ions, Mn^{2+} ions and/or non-ionic detergent.

27. The method according to claim 26 specifically used for producing radioactively or chemically labeled RNA probes and comprising an optional step of purifying the newly produced labeled RNA from the components of the reaction mixture.

28. The method according to claim 27, wherein said labeled RNAs are used as probes for Southern or Northern blot analyses after the optional purification step.

29. The method according to claim 27, wherein said labeled RNAs are used as probes for a fluorescent *in situ* hybridization analysis after the optional purification step.

30. The method according to claim 27, wherein said labeled RNAs are used as probes for a microarray analysis after the optional purification step.

31. A method for studying nucleic acid secondary structure, preferably RNA secondary structure comprising the steps of:

(a) providing nucleic acid target molecule, preferably RNA target molecule;

(b) contacting said target molecule with the protein or polypeptide defined in claim 1 or 12 under conditions sufficient for RNA synthesis in a mixture additionally comprising radioactively or chemically labeled nucleotides, so that single-stranded elements of said target RNA are copied by the polymerase of this invention;

(c) recovering and optionally purifying the newly produced labeled nucleic acid species from the reaction mixture;

- (d) using said labeled nucleic acid species as probes for microarray chip that comprises nucleic acid fragments of said target molecule;
- (e) interpreting data from the microarray analysis to deduce which parts of the target molecule are single-stranded; and optionally
- (f) building a model for the secondary or tertiary structure of the target molecule.

32. A method for studying nucleic acid-protein interactions, preferably RNA-protein interactions comprising the steps of:

- (a) providing a nucleic acid target and nucleic acid binding protein, preferably an RNA target and an RNA-binding protein;
- (b) contacting said target and the solution of said protein in an experimental mixture under conditions sufficient for target-protein interaction, and in a separate vessel, contacting said target with a control solution that lacks said protein.
- (c) contacting said experimental and control mixtures with the protein or polypeptide defined in claim 1 or 12 under conditions sufficient for RNA synthesis;
- (d) recovering and optionally purifying the newly produced labeled nucleic acid species from both reaction mixtures;
- (e) using the two sets of labeled nucleic acid species as probes for two identical microarray chips that comprise nucleic acid fragments of the target;
- (f) interpreting data from the two microarray analyses to deduce which parts of the target molecule are accessible for the RNA synthesis;
- (g) comparing the two data sets to determine the difference between target in experimental and control mixtures; and optionally
- (h) interpreting the difference between the two data sets as a model for nucleic acid-protein interactions

33. A method for producing RNA trigger molecules to induce RNA interference *in vivo* or *in vitro*, comprising the steps of:

- (a) providing RNA or DNA template;

(b) contacting said RNA or DNA template with the protein or polypeptide defined in claim 1 or 12 under conditions sufficient for RNA synthesis in a mixture comprising: nucleic acid template, protein defined in claim 1 or 12, nucleoside triphosphates, and optionally pH buffer, ammonium acetate, PEG, Mg^{2+} ions, Mn^{2+} ions and/or non-ionic detergent; and

(c) incubating the reaction mixture at temperature sufficient for RNA synthesis.

34. The method according to claim 33, wherein said RNA or DNA template originates from a cell or a virus.

35. The method according to claim 33, wherein said RNA template is provided by transcribing a DNA template with a DNA-dependent RNA polymerase, preferably derived from a bacteriophage selected from the group of T7, T3, and SP6 bacteriophages.

36. The method according to claims 33 or 35, wherein steps (a) and (b) are carried out at the same time or sequentially in the same reaction vessel.

37. A kit comprising the protein or polypeptide defined in claim 1 or 12.

38. The kit according to claim 37, wherein the kit further comprises additives necessary for a detectable level of RNA synthesis.

39. The kit according to claims 37 or 38 comprising nucleoside triphosphates in concentrations sufficient for RNA synthesis.

40. The kit according to any one of claims 37 to 39, wherein at least one nucleoside triphosphate is labeled with a radioactive isotope or is chemically modified.

41. The kit according to any one of claims 37 to 40, additionally comprising a standard nucleic acid preparation (or preparations) with characterized capacity to serve as a template (templates) for RNA synthesis by the protein or polypeptide defined in claim 1 or 12.